



Eromise Solânge Semedo Cardoso Varela

Licenciada em Engenharia Química e Biológica

Value and characterization of Cabo Verde Leguminosae plants as sources of new foods

Dissertação para obtenção do Grau de Mestre em
Tecnologia e Segurança Alimentar

Orientadores: Maria Manuel Romeiras, Professora Auxiliar, Instituto
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Preamble

The study whose results are presented in this dissertation was carried out at the Faculdade de Ciências e Tecnologia da Universidade Nova de Lisboa and Instituto Superior de Agronomia (ISA) da Universidade de Lisboa, within a research grant of the project *CVAgrobiodiversity - Climate changes and plant genetic resources: the overlooked potential of Cabo Verde's endemic flora*, funded by national funds of Fundação para a Ciência e a Tecnologia, I.P. (FCT)/MCTES and the Aga Khan Development Network (AKDN).

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This dissertation is based on the law (*n.º 2, art. 8.º, Decreto Lei n.º 388/70; República Portuguesa*), where most of the chapters are results of work for publication including work in collaboration. The author of the Master dissertation declares that she participated in the conception and execution of the experimental analysis, discussion, and preparation of the work for publication.

Abstract

Legume species are important food sources to prevent starvation, under-, and malnutrition; they also play a crucial role in sustainable agriculture in the tropical dry islands of Cabo Verde. In order to improve the knowledge of the heritage of plant genetic resources in Cabo Verde, namely of the Leguminosae family, this study had three main goals: i) to provide a checklist of Leguminosae taxa used as food; ii) to investigate which legume species are consumed and traded in local markets and to compare species for their chemical composition and nutritional value; and iii) to discuss aspects concerning the agronomic value, sustainable use of legumes, and their potential contribution to food security in this archipelago. Results revealed that 15 Leguminosae species are used as food and all but one is cultivated. Five of these species are widely consumed/traded in local markets, namely: *Cajanus cajan*, *Lablab purpureus*, *Phaseolus lunatus*, *Phaseolus vulgaris*, and *Vigna unguiculata*. Regarding the nutritional value, *Lablab purpureus*, *Phaseolus vulgaris* and *Vigna unguiculata* present about 23% of protein. *Cajanus cajan* and *Lablab purpureus* showed the highest antioxidant capacities, fibre and phenolic contents; *Phaseolus vulgaris* showed the highest ash and moisture contents. The highest mineral content was found for the following elements and species: Cu, in *Cajanus cajan*; Mg, and Mn, in *Lablab purpureus*; K, Ca, P, and Fe in *Phaseolus vulgaris*; and Mg, S, and Zn in *Vigna unguiculata*. The role of these species as sources of nutrients for food security is highlighted, and the native ones (*Lablab purpureus* and *Vigna unguiculata*) stand-out as particularly well-adapted to the adverse climate of these islands. The present study allowed concluding that the conservation and sustainable use of these plant genetic resources can contribute to the reduction of hunger and poverty, thus meeting some challenges of the Sustainable Development Goals (zero hunger, good health and well-being and climatic action).

Key-words: Oceanic Islands; Middle Income Countries; diversity of legume species; phenolic contents; mineral content; nutritional composition; Agronomic value.

Resumo

As espécies de leguminosas são importantes fontes alimentares para evitar a fome, a subnutrição e a má nutrição. Desempenham um papel crucial na agricultura sustentável das ilhas tropicais secas de Cabo Verde. A fim de melhorar o conhecimento do património de recursos genéticos vegetais em Cabo Verde, nomeadamente da família Leguminosae, este estudo teve três objetivos principais: i) fornecer uma *checklist* dos *taxa* Leguminosae utilizados como alimentos; ii) investigar que espécies de leguminosas são comercializadas nos mercados locais e avaliar a sua composição química, valor nutricional, conteúdo fenólico e actividade antioxidante; iii) discutir aspectos relativos ao valor agronómico, utilização sustentável das leguminosas, e a sua potencial contribuição para a segurança alimentar neste arquipélago. Os resultados revelaram que cinco espécies são amplamente consumidas/comercializadas nos mercados locais, nomeadamente: *Cajanus cajan*, *Lablab purpureus*, *Phaseolus lunatus*, *Phaseolus vulgaris* e *Vigna unguiculata*. As espécies *Lablab purpureus*, *Phaseolus vulgaris* e *Vigna unguiculata* apresentaram um teor em proteína de cerca de 23%. As espécies *Cajanus cajan* e *Lablab purpureus* mostraram as mais altas capacidades antioxidantes e maior conteúdo em fibras e compostos fenólicos; enquanto que, o *Phaseolus vulgaris* apresentou maiores teores em cinza e de humidade. Os valores mais elevados de minerais foram encontrados para os seguintes elementos e espécies: Cu, em *Cajanus cajan*; Mg e Mn em *Lablab purpureus*; K, Ca, P e Fe em *Phaseolus vulgaris*, e Mg, S e Zn em *Vigna unguiculata*. O papel destas espécies como fonte de nutrientes importantes para a segurança alimentar fica enfatizado. O trabalho efetuado permitiu concluir que a conservação e utilização sustentável destes recursos genéticos vegetais pode contribuir para a redução da fome e da pobreza, respondendo assim a alguns desafios dos Objetivos de Desenvolvimento Sustentável.

Palavras-chave: Ilhas oceânicas; Países de Rendimento Médio; diversidade das espécies de leguminosas; conteúdo fenólicos; conteúdo mineral; composição nutricional; valor agronómico.

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Acronyms and Abbreviations

AAE - Ascorbic acid equivalents

DPPH - 2,2-diphenyl-1-picrylhydrazyl

EFSA - European Food Safety Authority

FAO - Food and Agriculture Organization

GAE - Gallic acid equivalents

ICP-OES - Inductively coupled plasma - optical emission spectrometry

IUCN - International Union for Conservation of Nature

Ma- Megaannum (one million years)

RS - Resistant starch

SCFA - Short-chain fatty acids

SD - Standard deviation

PC - Principal Component

PCA - Principal Component Analysis

PDCAAS - Protein Digestibility Corrected Amino Acid Score

UA - Univariate Analysis

CHAPTER I.

1. Introduction

Since ancient times, mankind has been able to recognize the relationship between food and health. Hippocrates “the father of medicine”, more than two millennia ago stated “let's food be your medicine and let medicine be your food” (Shultes, 1978). However, there are some foods that in addition to providing basic nutrients and energy, also provide biologically active components that can improve the physical and /or mental health of the consumer, preventing or reducing the risk of developing certain diseases or health problems. These foods are known as “functional foods” (Bennett et al., 2015). Reasons such as increasing human health awareness, advance of knowledge and technology in the food industry and medicine and branches of the natural sciences studying the relation between nutrition and health have led to the popularization of foods with proven health effects (Vukasović, 2017). Nowadays, the investment on nutrition and research has increased. Human nutrition has always been very important, not only to satisfy dietary needs, but also as a strategy to protect and /or reduce the evolution of various diseases because of a weak, unbalanced or excessive intake of nutrients (Muzquiz et al., 2012).

1.1. Leguminosae

Leguminosae (legumes) are the third largest plant family, with approximately 730 genera and 19,400 species, and they are found throughout the world in all biomes. Belonging to the Fabaceae family, leguminous seeds, also known as pulses, constitute a worldwide staple food, being surpassed only by *Poaceae* (the grasses) in agricultural and economic importance, representing about 15% (270 a 300 million hectares) of the world arable land. The name 'Fabaceae' comes from the defunct genus *Faba*, "faba" comes from Latin and appears to simply mean "bean" (Albala, 2007).

Although sometimes used interchangeably as synonyms, the terms “legumes,” “pulses,” and “beans” have distinct meanings. There are commonly referred to as “pulse”, a term probably deriving from the Latin *puls* meaning pottage, while the term “legume” originates from the Latin *legumen* describing seeds harvested from pods (Dilis & Trichopoulou, 2009). A legume refers to any plant from the Fabaceae family that would include its leaves, stems, and pods

(Yusuf & Abdullahi, 2019). The legumes can be separated into two classes: oilseeds such as soybeans and peanuts, which are grown for both their protein and oil content, and grain legumes including common beans, lima beans, cowpeas, fava beans, lentils, chickpeas, common peas which are grown primarily as a protein source. It is estimated that there are ca. 13,000 species of the legumes, but only about 20 are commonly consumed by humans (Geil & Anderson, 1994; Maphosa & Jideani, 2017). Fabaceae family only, is responsible for the greatest part of the diversity of the order Fabales. Attempts to estimate the age of legumes and diversification in the family, based on molecular markers to reconstitute their phylogeny, have been published in recent years (Maphosa & Jideani, 2017). Wikström et al. (2001) estimated an age for Fabaceae of 74-79 Ma, Moore et al. (2010) suggested ages of 107-100 Ma and Bell et al. (2010) ages of 107-91 Ma. There are still controversies about the origin of plants belonging to this family, however fossil records found in North America, Europe, Africa and Asia indicate that pulses emerged at least 60 million years ago, due to the rapid desertification even that happened in the World. Domestication of pulses in the Old World goes back to Neolithic times and was approximately contemporary to that of the cereals (De Ron, 2015). These plants are found in practically all plant formations of the planet, although the current centre of endemism of the family is the Neotropical region (Lavin et al., 2004). According to the Food and Agriculture Organization (FAO) definition, the term “pulses” includes only dry edible seeds with low fat content, therefore beans, peas or fava beans, that are harvested while still green are not included in this definition (FAO, 1994). Because of their importance, the year 2016 has been declared by FAO as the International Year of Pulses (UN, 2013) and in 2019, United Nations designated February 10th as World Pulses Day. This staple food has been a relevant constituent in human food, important not only to combat malnutrition and ensure food security, as they are a critical and relatively accessible source of valuable nutrients, but also to reduce poverty and improve human health, since their regular consumption can help prevent and/or manage some growing public health concerns associated with an urban life-style (Akibode & Maredia, 2011; Singh 2017). They are also an important part of a sustainable cropping system, fixing nitrogen to the soil, reducing the need for water, and improving soil health (Karkanis et al., 2018; Siegel & Fawcett, 1976). Many pulse species are drought-tolerant, carrying broad genetic diversity from which climate-resilient varieties can be selected and/or bred making them an ideal crop for dryland regions (Saikia et al., 2018). Dry edible beans have been playing a prominent role in the diets of many countries and communities, with remarkable nutritious value and many health benefits. Early men grew and used dried beans as a staple food from early biblical times, long before modern nutrition

researchers transmitted their health benefits (Messina, 2014). The figure 1.1 shows the seed structure of a bean, with its constituent parts.

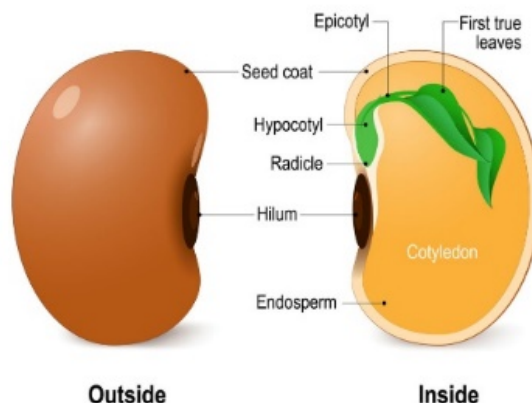


Figure 1.1-Seed structure of beans (Retrieved from: <http://naturejournals.org/index.php/environments/classification/fruit-seeds/#author>)

1.1.1. Composition and nutritional value of dry beans

Although dried beans all belong to the same botanical family, its size, colour, shape and flavour differ greatly between varieties (Geil & Anderson, 1994). However, despite the plethora of organoleptic differences, beans are surprisingly similar in nutrient composition. Nonetheless, the nutritional composition varies according to the type of beans, place of production, environmental factors, climate as well as the type of soil on which they are grown (Fabbri & Crosby, 2016; Rebello et al., 2014). Their nutrient profile fits with the dietary needs of people of all ages (Ordovas et al., 2018). Some recent studies have pointed that pulse by-products, seed coats and pods for instance, are far from being utilised as potential sources of ingredients with food application at their full potential (Singh et al., 2017). It is known that processing is essential to make food easier to digest and increase energy and nutrient availability (Weaver et al., 2014). Food processing methods, from soaking to germination, fermentation, and cooking (boiling) greatly influences beans nutritional value (Nkhata et al., 2018). Processing of dry beans keeps functional ingredients and preserves their nutritional characteristics, influencing the bioavailability, utilization of nutrients, improving palatability thus ensuring a positive impact on consumers' health (Ibrahim et al., 2002). Perhaps one of the most known nutritional characteristics of beans is their high protein content, which is 2 to 3 times higher than that of cereal grains (Siddiq et al., 2010). Dry beans are

grouped along with fish, meat, nuts, and eggs as high sources of protein (Hess & Slavin, 2016). Dry beans also include in their constitution several types of prebiotics, including resistant starch (RS) and the fructooligosaccharides, stachyose and raffinose (Geil & Anderson, 1994). Prebiotics play an important role in intestinal flora, serving as a substrate for bacterial fermentation in the human intestine, thus influencing microorganisms of the gastrointestinal tract and intestinal metabolism (Davani-Davari et al., 2019). In terms of calories, a portion of 100 grams of raw beans provides an average of 345-350 kcal, whilst the same portion (approximately ½ cup) of cooked dry beans provides 110-143 kcal. The caloric value for raw beans is given mainly from carbohydrates, where starch is the predominant component (Noah et al., 1998). The caloric value of beans varies with moisture content, where the lower caloric species are also higher in moisture (Geil & Anderson, 1994).

Proteins are an important macronutrient family. They are complex polymers made from twenty basic building blocks called amino acids, connected through peptide bonds. Of the 20 amino acids present in proteins, 9 are considered nutritionally indispensable (essential) in adult humans because the body is not able to synthesize their carbon skeletons. These nine are: histidine, isoleucine, leucine, lysine, methionine, threonine, tryptophan and phenylamine (Pujolà et al., 2007; Watford & Wu, 2018). Dry beans are a good source of protein, being composed by an average of 21-25% crude proteins, soybeans being the exception, with approximately 34% protein content (Sathe, 2002). Despite being a great source of protein, beans are considered of low biological value, since they have limitations of sulfur amino acids, such as methionine and cysteine, as well as of tryptophan (Rebello et al., 2014). In order to compensate for this limitation, it is advisable to combine beans with other foods, cereals for instance, in order to have a more complete diet (Mariotti & Gardner, 2019). On the other hand, beans contain significant amounts of lysine, an amino acid normally found in low levels in cereal proteins (Alonso et al., 2010). The nutritional quality of proteins is not only dictated by their amino acids composition, but also by their digestibility in the gastrointestinal tract, which in turn depends on their structure and enzymatic accessibility (Achinewhu & Hewitt, 1979). Protein digestibility, which can be calculated using the Protein Digestibility Corrected Amino Acid Score (PDCAAS), is one widely used parameter to estimate protein quality (Schaafsma, 2000). Regarding beans, raw beans proteins are much less available than the proteins of cooked beans, as food processing may affect the bioavailability and bioactivity of pulse proteins and peptides (Margier et al., 2018).

Carbohydrates are one of the three macronutrients in the human diet, along with fat and proteins. Carbohydrates are known as saccharides or sugars and their main function is to provide energy (Slavin & Carlson, 2014). Most abundant carbohydrates in legume seeds are starch and non-starch polysaccharides (dietary fibre), with smaller but significant amounts of oligosaccharides (Lunn & Buttriss, 2007). The soluble carbohydrate fraction of dry beans contains monosaccharides (glucose, fructose, and galactose), disaccharides (sucrose), and raffinose family of oligosaccharides (Guillon & Champ, 2002). Those oligosaccharides, along with other beneficial compounds, are fermented by the colonic microflora producing beneficial short-chain fatty acids (SCFA), account for nutritional and functional value attributed to pulses (Campbell et al., 1997). The total carbohydrate in dry beans is high, ranging from 60 - 70%. The most abundant carbohydrate is starch, ranging from 31.5-53.6 %, and the total sugars (mono and oligosaccharides) constitute a reduced portion of total carbohydrates in beans (Ofuya & Akhidue, 2005). The resistant starch content of beans is much higher than in commonly consumed grains, most likely because of their high ratio of amylose to amylopectin (Messina, 2014; Tosh & Yada, 2010).

Dietary fibre are plant-based carbohydrates that are not digested in the small intestine and so reaches the large intestine or colon (Dhingra et al., 2012). Crude fibre, consists of cellulose, hemicelluloses, lignin and pectin and is known to promote beneficial physiological effects for human health (Iqbal et al., 2013; Tosh & Yada, 2010). Dry beans contain a substantial amount of carbohydrate as fibre in the form of cellulose and hemicellulose. Fibre-rich foods may help to promote satiety and are linked to weight regulation (McCrory et al., 2010). To tackle the low intake of dietary fibre, some countries like Canada, which are major world producers and exporters of leguminosae, but whole seeds have a low market value, mill and fractionate leguminosae seeds to isolate important components in dietary fiber for incorporation in commercial foods, thus enriching fibre content and acting as a functional ingredient (Tosh and Yada, 2010). The fibre in beans also aids in normal functioning of the gastrointestinal tract due to its bulking properties, hydration capacity, binding properties and fermentability, especially in comparison with cereals (Rebello et al., 2014). The European Food Safety Authority (EFSA) recommended an intake of 25 grams of fiber per day for adults. For children the recommended intake depends on the age, being 10g/day (from 1 to 3 years), 14 g/day (from 4 to 6 years), 16 g/day (from 7 to 10 years), 19 g/day (from 11 to 14 years) and 21 g/day (15 to 17 years) (EFSA, 2010).

From a nutritional point of view, beans are considered one of the most important sources of minerals, in fact, they belong to the group of crops that easily accumulate metals from soil (Lovato et al., 2018). Beans contain minerals, such as copper (Cu), iron (Fe), magnesium (Mg), manganese (Mn), phosphorous (P), potassium (K) and zinc (Zn). Legumes, in general, offer a diversity of micronutrients exceeding or complementing the profiles of cereals (Polak et al., 2015; Tardy et al., 2020). The world population, especially the poorest countries, is at risk for micronutrient intake, recent studies showing that Fe deficiency is the most prevalent micronutrient problem in the world and that deficiencies in several other minerals, such as Zn, Mg and Ca, are also a problem in many parts of the world (Barba & Feliciano, 2002). Some studies suggest the use of biofortified beans as a potential solution to tackle this problem. However, the presence of antinutrients often is associated with a reduction on the bioavailability of minerals (Caproni et al., 2020). There are studies focused in reducing the content of components such as phytic acid in order to improve minerals absorption (Celmeli et al., 2018; Tako et al., 2015).

Fat is a very diverse group of hydrophobic compounds, including free fatty acids, mono, di and triacyl glycerol, phospholipids, sterols, esters, glycolipids, and lipoproteins (Ofuya & Akhidue, 2005). The total lipids content of beans is very low, ranging from 1.0 – 4.99 %. There are some factors, which influence the lipid content in leguminosae: origin, variety, climate and location of species (Barampama & Simard, 1993). The harvest time has a strong influence on the composition of fatty acids (Iqbal et al., 2013).

Several water soluble vitamins, namely vitamins, B1 (thiamine), B9 (folic acid), B2 (riboflavin), B3 (niacin) and vitamin B6 (pyroxidine) were identified in dry beans (Celmeli et al., 2018). There are investigations that have shown that the presence of B9 and B6 vitamins can be useful in reducing the risk of cardiovascular diseases. B9 is known for reducing the risk of neural tube defects in new-borns and it's widely recommended for pregnant women (Grosse & Collins, 2007; Simmons, 2013). Vitamin C in dried beans was found in relatively considerable amounts, however 70-100% has lost through cooking. Little to no fat-soluble vitamins was found in dry beans due to the low level of lipids (Baardseth et al., 2010).

1.1.2. Health benefits of dry beans

Phytochemicals of beans have a great potential as a functional and nutraceutical ingredient possessing antioxidant, anti-inflammatory, anti-hypertensive, anti-atherosclerotic, antitumor and antiaging properties (Ganesan & Xu, 2017; Messina, 2014). The incorporation of beans in

the diet has been associated with improved lipid parameters and colic function, as well as decreased risks of developing pathologies such as diabetes, cardiovascular (Messina, 2014; Singh et al., 2017). It has been reported that the protective effects of dry beans in disease prevention, such as against cancer, may not be entirely associated to dietary fiber, but to phenolics and other non-nutritive compounds (Siddiq et al., 2010). The color of the bean coat is linked with the antioxidant capacity, colored beans (red, brown or black) possessing greater antioxidant activity than white beans (Bressani, 1993). Researchers have found that the inclusion of beans in diets attenuates postprandial insulin and moderately enhances postprandial antioxidant endpoints in adults with metabolic syndrome (Reverri et al., 2015).

1.1.3. Antinutrients

Some plant compounds can interfere with the body's ability to absorb essential nutrients. Those substances are not usually a concern, but during periods of malnutrition, or among people with low dietary diversification, they may constitute a problem. These compounds are known as antinutrients and include, among others, protease inhibitors, alpha-amylase inhibitors, phytic acid, lectins, glycosides cyanides, tannins and oxalates (Carbas et al., 2020). The presence of anti-nutritional factors in beans is shown to be reduced at varying degrees, according with the chosen food preparation method. Thermal treatment, especially moist heat induces changes in the protein structure that may inactivate antinutritional factors thus increasing the digestibility and biological value of the bean's proteins (Urbano et al., 2000). Soaking has been also found to decrease phytate, protease inhibitors, lectins, tannins and calcium oxalate. Sprouting the legumes lead to the degradation of antinutrients such as phytate, lectins and protease inhibitors (Samtiya et al., 2020). Other method, although not so common in the West, is the fermentation of grain legumes, which is also a very effective method of reducing antinutrients and increasing the nutrient absorption. Some of these methods are often combined, enhancing, thus, their, antinutrient reduction potential (Difo et al., 2015). Data shows that the amount and profile of such antinutrients can vary depending on climate and further environmental conditions, as well as according to the species (Popova & Mihaylova, 2019). It is important, however, to understand that the effects of individual components studied in isolation are often not the same when these components are consumed after processin and combined with other foods, as normally occurs in the diet (Carbas et al., 2020).

1.1.4 Bioactive compounds of beans

Phenolic compounds are present mainly in the seed coat of beans, while cotyledons and testa may also contain these nutraceutical ingredients but only in small amounts (De Mejia et al., 1999). The phenolic compounds commonly present in the seeds are flavones, monomers, and oligomers of flavanols, flavanones, isoflavonoids, anthocyanins, chalcones, and dihydrochalcones. However, the phenolic acids and non-flavonoid phenolic compounds (hydroxybenzoic and hydroxycinnamic acid) are mainly found in cotyledons of the bean, while condensed tannins, proanthocyanidins and anthocyanins reside mainly in the testa (Suárez-Martínez et al., 2016; Troszynska & Ciska, 2002). Polyphenolic compounds can largely influence seed coats' colour patterns due to diversification and variability in the composition of procyanidins, flavonol glycosides and anthocyanidins (Kilonzi et al., 2017). Darker beans normally have the highest anthocyanins concentrations whereas light yellow or pink spot of the seed coat are generally based on the presence of condensed tannins. The Table 1.1 shows the polyphenolic content of different types of beans (Ganesan & Xu, 2017).

Table 1.1: Polyphenols identified in some types of beans (Ganesan & Xu, 2017)

Beans Name	Polyphenol Class	Major compounds
Dark beans	Flavonoids, Flavones; Phenolic acids	Cyanidin 3-O-glucoside; Pelargonidin 3-O-glucoside; caffeic acid
Brazilian beans	Flavonoids	Chrysin; Quercetin; Kaempferol
Red beans	Flavonoids; phenolic acids	kaempferol, astragalin, (+)-catechin, <i>p</i> -coumaric acid, hydroxybenzoic acid, ferulic acid, caffeic acid
White bean	Flavonoids; phenolic acids	(+)-Catechin, <i>p</i> -coumaric acid, hydroxybenzoic acid, ferulic acid, caffeic acid

The level of total phenolics is influenced not only by genetic factors, but also by growing location (environmental factors) and genotype (Kilonzi et al., 2017).

Although aerial parts of beans are edible, their popularity as food items is low due to lack of good flavour, texture or just because they are not commonly included in the culinary repertoire of many cultures. In Africa, however, bean leaves are a regular part of local diets in many countries serving both as a vegetable addition to recipes as ingredient in the preparation of traditional medicinal (Goswami & Hitendra, 2017). Regarding polyphenolic content,

studies have showed that bean leaves present higher content of polyphenolic substances exhibiting thus stronger antioxidant than the seeds (Ko et al., 2014). After the harvest of edible beans, a significant amount of leaves is generated, which are treated as waste, fertilizer or used as animal feed. The functional and nutraceutical properties have not yet been properly studied.

1.1.5. Beans worldwide production and consumption

It has been shown through studies that changes in consumption patterns directly affects nutritional outcomes. The results of changing consumption patterns are important for policy makers because they are concerned with food and nutrition security. Consumption patterns are affected by factors such as average incomes, food cost, urbanisation, globalisation, demographic shifts, market exchange policies, transportation and changing consumer tastes and preference (Kearney, 2010). Despite the proven benefits, the *per capita* consumption of pulses has steadily declined in both developed and developing countries. Actually, the inhabitants of several African countries support their diets on cereal staple foods, subsisting on a mono carbohydrate diet (for instance, rice or maize) and lacking an adequate protein supply (Huang & Drescher, 2015). Most crop improvement efforts of the "Green Revolution" emphasized the cereals rather than pulses. In some areas, total beans production has actually declined in favour of the cereal grains, even though many developing nations suffer from a chronic protein shortage. Fortunately, this situation is now slowly being reversed (Figure 1.2). On a recent study (OECD, 2019), India ranked the highest in dry bean consumption, followed by Myanmar and Brazil. When referring to *per capita* consumption, however, Niger, Myanmar and Burkina Faso recorded the highest levels. The lowest ranked countries in that category are Turkmenistan, Guinea and Sierra Leone. Beans are widely consumed around the World in the form of fresh (green) or processed (dried or cooked) grains, forming part of the basis of traditional local recipes, representing 50% of legumes consumed as food (Di Bella et al., 2016; Huang & Drescher, 2015).

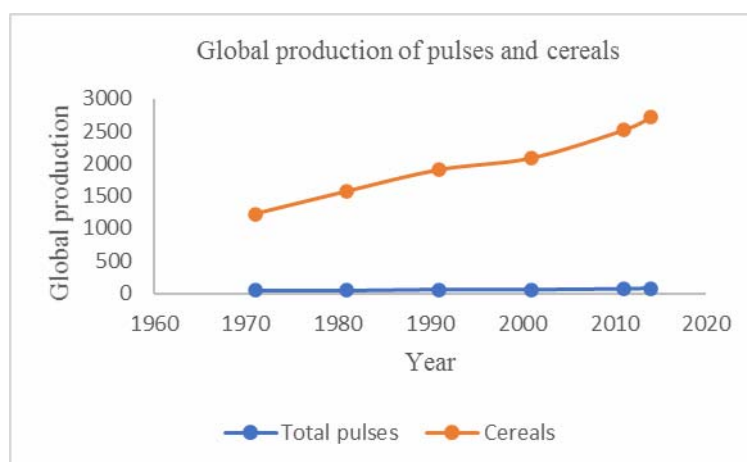


Figure 1.2: Global production (million tonnes) of pulses and cereals in different years (FAOSTAT, 2019)

1.2. Cabo Verde Leguminosae species: Contribution to food security

Agricultural development is imperative to improve food security and nutrition (HLPE, 2016). Increasing the quantity and diversity of food will provide the primary source of income for many people, which is particularly important in low- and middle-income countries, namely those in the African continent (Varela et al., 2020). The FAO recently assessed the list of countries undergoing food emergency and in need of external assistance for food, where 34 out of 45 countries are in Africa (FAO, 2020). The effects of the COVID-19 pandemic, particularly through the loss of income and jobs related to confinement measures, severely aggravated global food security conditions (Otekunrin et al., 2020). These are even more worrying in countries where food security was already a major concern, such as the tropical dry islands of Cabo Verde where the agriculture sector is extremely limited by natural constraints such as drought period, poor soils, scarcity of cropland and low technological level of implementation (Varela et al., 2020).

The Cabo Verde archipelago comprises the southernmost islands of Macaronesia (i.e., Azores, Madeira, Savage Islands, Canary Islands, and Cabo Verde) and it is located in the Sahelian arid and semiarid regions in close proximity to the western African coast (Neto et al., 2020). Cabo Verde consists of nine inhabited islands grouped in Northern Islands (São Nicolau, São Vicente and Santo Antão), Eastern Islands (Sal, Boavista and Maio) and Southern Islands (Santiago, Fogo and Brava). The vascular flora comprises ca. 740 vascular plant taxa, of which ca. 92 are endemic (Romeiras et al., 2020). In a recent conservation assessment based on the International Union for Conservation of Nature (IUCN) Red list criteria, ca. 78% of the endemic plant species were considered threatened, mostly as a

consequence of the growing habitat degradation, human disturbance (e.g. intentional use for agriculture or traditional uses) and introduction of exotic species since the beginning of the islands' colonization (Romeiras et al., 2016).

Cabo Verde was the first tropical archipelago colonized by Europeans, in 1462, and due to its geographic location in the mid-Atlantic Ocean, it became an important hub for trans-Atlantic trade routes (Albuquerque, 1991). Particularly since the 16th century, these islands played an important role as a centre of dissemination and acclimatization of tropical crop species of key economic importance, prior to their cultivation in other regions (Havik et al., 2018). Also, during human settlement, the introduction of useful plants was of chief importance to change the present-day composition of the archipelago's flora, which comprises a great number of exotic taxa (Romeiras et al., 2011). Historical contingencies, namely the location of the main harbours (i.e., Mindelo, in São Vicente, and Cidade da Praia, in Santiago) and the establishment of the first settlements greatly impacted the knowledge of local flora and the botanical explorations in this archipelago (Romeiras et al., 2020).

Although several botanical explorations were performed in this archipelago since the late 18th century (Francisco-Ortega et al., 2015; Rico et al., 2017; Romeiras et al., 2014), only in 1908 was the first Agronomic Mission carried out, by P. de Lemos, Pereira da Cunha and A. Costa Andrade. In 1935, the French botanist Auguste Chevalier (1935) published a seminal work "*Les Iles du Cap Vert. Géographie, biogéographie, agriculture. Flore de l'Archipel*" covering the vast majority of the plant collections made in these islands since the late 18th century. During the colonial period (1915–1974), Grandvaux Barbosa assembled the largest plant collection, and published the first comprehensive study of the territory's agriculture (Teixeira & Barbosa, 1958). This study stressed that the main rainfed crops of Cabo Verde were maize (*Zea mays* L.) and several bean species (i.e., *Cajanus cajan* (L.) Huth, *Lablab purpureus* (L.) Sweet, *Phaseolus vulgaris* L., *Phaseolus lunatus* L. and *Vigna unguiculata* (L.) Walp.). These species are still the basis of the country's diet, being the most emblematic dish of Cabo Verde called "cachupa", which is cooked with different varieties of beans and maize. These crops are produced through rainfed subsistence farming, whereas irrigated crops, such as sugarcane and tomatoes, are mostly grown for commercial purposes. Monteiro et al. (2020) reported that Santiago has the largest area used for agriculture (52.5%), followed by Santo Antão (16%) and Fogo (15.8%). These islands are better for agriculture than the others, because they have a complex variety of microclimates, ranging from more humid zones in mountain regions of Santiago (Pico da Antónia) and Santo Antão (e.g. Ribeira do Paúl), to volcanic areas in Fogo, which reaches almost 3000 meters, or to lowland arid areas that experience the scourge of

long-lasting droughts. During the 20th century, anthropogenic activities caused enormous damage and, particularly in humid and sub-humid areas on the N and NE slopes above 400 m, natural vegetation was gradually cut and destroyed and replaced by crop species (Norder et al., 2020). Nevertheless, several conservation actions were undertaken over the last two decades by Cabo Verde authorities, in particular a system of Protected Areas to safeguard the natural heritage of the archipelago (MAAP, 2004; Romeiras et al., 2016).

Since the Convention on Biological Diversity (see: <https://www.cbd.int/intro/>) several steps were taken at international levels to address specific biodiversity issues, such as agrobiodiversity and sharing benefits associated with the exploitation of the plant genetic resources. Despite the key role played by the plant genetic resources in the Cabo Verdean agriculture, they are still poorly known and there is no clear understanding of which ones are relevant to the improvement of crop species, as well as their potential capacity to adapt to climatic and environmental changes. Particularly, pulses (i.e., edible seeds of plants of the legume family) have a long history as staple crops for smallholder farmers in semi-arid tropical areas of sub-Saharan Africa (Snapp et al., 2019) and, as stated above, are among the most important crops of Cabo Verde (Monteiro et al., 2020; Teixeira & Barbosa, 1958).

1.2.1. Pulse species cultivated in Cabo Verde

After the cultivation of corn, beans represent one of the most important crops, both for the area it occupies and for the total productions it reaches during the harvest. Beans are widely consumed by the population, where the leaves are used as fodder for domestic animals. *Cajanus cajan*, *Lablab purpureus*, *Vigna unguiculata*, *Phaseolus lunatus* and *Phaseolus vulgaris* are five species of beans consumed in Cabo Verde.

Cajanus cajan (L) Millsp is usually known in english as Pigeon Pea or Cajan Pea, the last designation being most used by Americans. In Mozambique it is known as Cowpea, already in Cabo Verde (Santiago Islands) it is known as *feijão congo* (congo beans) and is an introduced species (Teixeira & Barbosa, 1958). The center of origin is most likely Asia, where it then traveled to East Africa through the slave trade to the American continent. It is one of the most important dryland pulses in the tropics and semi-arid regions (Pal et al., 2011). In some countries like Trinidad and Tobago they use *Cajanus cajan* leaves to treat food poisoning problems such as colic and constipation, still in the eastern part (India) they use the leaves, seeds and young stems in the treatment of gingivitis and stomatitis (Lans,

2007). This species plays a crucial role particularly in the more rugged islands, due to its resistance to winds, and because it is consumed green and dry. In addition to feeding it is used in cattle fodder, and the parts resulting from pruning serve as woody material for fuel to replace the butane gas.

Lablab purpureus L. Sweet is commonly known as stone beans (feijão pedra em Cabo Verde), is a native species of Cabo Verde, which grows in the tropical and subtropical regions of Asia and Africa. This species has the potential in improving human food and animal feed as a vegetable, pulse and/or forage crop, and is important in tropical agricultural systems, since it grows in various environments and is drought tolerant (Kimani et al., 2019).

Phaseolus lunatus L. is commonly known as lima beans or butter beans. This is an introduced species in Cabo Verde, very appreciated and consumed in the archipelago, known in Cabo Verde as fava ou feijão lima. From this variety may appear some that are toxic differentiating in color and organoleptic characteristics (Teixeira & Barbosa, 1958). *Phaseolus Lunatus* is a popular source of protein in several countries of Africa (Purwanti & Fauzi, 2019). In the same pod, it is possible to find colors red or yellow-brown and white.

Phaseolus vulgaris L., commonly known as "common bean ", is one of the most important leguminosae for human consumption in the World. In Cabo Verde this species is known as feijão carioca ou sapatinha. *Phaseolus vulgaris* had its origin in America, where it then moved to Europe and from Portugal to Cabo Verde (Teixeira & Barbosa, 1958).

Vigna unguiculata (L) Walp, commonly known as Cowpea, is throughout the tropics and subtropics. Is a native species of Cabo Verde and an important crop in Africa, but is also grown in Brazil, India, Southeast Asia, and the United States (Allen, 2013). In Cabo Verde is known as bongolon, is widely sown and consumed in human food and as fodder (Teixeira & Barbosa, 1958). Cowpea play an important role in agronomic, environmental and economic terms, contributing to the improvement of diets and agricultural yields. It is highly valued for its nutritional value (low lipid content and high protein value), which is closely linked to the prevention of cardiovascular and metabolic diseases. This crop is consumed from leaves, green pods, green beans and ripe beans (Gonçalves et al., 2016).

1.3. Aims

In Cabo Verde Islands, local populations detain a large knowledge on the uses and properties of many native flora, but there is still a lack in scientific knowledge about the alimentary and chemical properties, particularly of native plant genetic resources. In order to improve the knowledge of the heritage of genetic resources of the Leguminosae family in Cabo Verde and to provide a deeper understanding of the importance and value of legume crops grown in this archipelago, as well as to give insight into the importance of bean leaves in the food base, this study had four main general objectives:

- 1) Provide a checklist of Leguminosae taxa used as food, and new data on their native distribution (archipelago and worldwide), common names, and other uses (i.e. forage and medicinal).
- 2) Investigate the legume species most consumed and traded in Santiago markets
- 3) Perform a physical-chemical and nutritional characterization of the most traded legume species
- 4) Discuss the agronomic value and the contribution of beans to food security in Cabo Verde

To achieve these general objectives, the following specific objectives were defined:

- Application of questionnaire to sellers in different markets of Santiago islands, in order to understand the consumption of beans, the importance of legumes for the Cape Verdean population, as well the frequency of demand;
- Studying the physical characteristics (color and size of the seeds) of different species;
- Determination of the chemical composition (mineral, ash, moisture, crude fibre and protein contents) of different species;
- Determination of the phenolic compounds and antioxidant capacity of the leaves of the different species.

CHAPTER II

2. Material and Methods

2.1. Studied species

Data on the food Leguminosae species known in Cabo Verde were obtained through a comprehensive review of the best knowledge currently available. The baseline data for the present study was gathered from Herbarium collections [Herbarium of Instituto Superior de Agronomia of the University of Lisbon, João de Carvalho e Vasconcellos (LISI); Herbarium of Instituto de Investigação Científica Tropical (IICT/ULisboa), University of Lisbon (LISC); Herbarium of Museu Nacional de História Natural e da Ciência, University of Lisbon (LISU); Herbarium of University of Coimbra (COI); Herbarium of Royal Botanic Gardens, Kew (K); Herbarium of Natural History Museum (BM); and Herbarium of Meise Botanic Garden (BR)]. Moreover, scientific publications (Barbosa, 1961; Duarte et al., 1996; Morrone, 2002; Romeiras et al., 2011; Teixeira & Barbosa, 1958; Varela, 2001) and online databases [e.g., Plants of the World Online (POWO, 2020), PROTA - Plant Resources of Tropical Africa (PROTA, 2020), IPNI - International Plant Names Index (IPNI, 2020) and the International Legume Database and Information Service (ILDIS, 2020)] were also accessed for information on taxonomic data, native distribution and cultivation status. We then constructed a comprehensive database including the scientific name of each species, English common names, native status in Cabo Verde, native distribution, habit and distribution in Cabo Verde.

2.2. Sampling

The field surveys (Supplementary Data) were performed between 2018 and 2019 on Santiago Island. This island is the largest one in the Cabo Verde archipelago, and also the one with the largest population, about a fifth of the inhabitants live on this island (INE, 2018a). The climate is characteristically hot and semi-arid, with the rainy season from August to October; September being the wettest month whilst the annual average temperatures attain the maximum of 25 °C (Neto et al., 2020). Surveys were made in the main trade markets of Santiago Island (Assomada, Calheta, Praia, São Domingos, São Jorge dos Órgãos, São

Salvador do Mundo and Tarrafal) in order to identify the most cultivated and traded bean species of Cabo Verde. Data on trade species, including their origin and availability were also obtained during the field surveys. Five species are widely consumed/traded in Santiago markets, namely: *Cajanus cajan*, *Lablab purpureus*, *Phaseolus lunatus*, *Phaseolus vulgaris*, and *Vigna unguiculata* (Figure 2.1 and Figure 2.2).



Figure 2.1: Studied bean species from Santiago Island. A. Field beans in the Assomada region, with details of *Cajanus cajan* (A.1) and *Vigna unguiculata* (A.2). B. The two main Santiago markets showing different beans sold in Assomada market (B.1) and Cidade da Praia municipal market (B.2). C. Details of the studied bean species: *Cajanus cajan* (C.1); *Lablab purpureus* (C.2); *Phaseolus lunatus* (C.3); *Phaseolus vulgaris* (C.4); and *Vigna unguiculata* (C.5).

Beans (dry seeds) from different markets and species were then suitably packaged and shipped by air to Portugal. Once in the laboratory (Lab. facilities of NOVA School of Science

and Technology, Caparica, Portugal), these beans were subjected to physicochemical analysis. A portion of each of the collected samples were germinated under controlled conditions in a greenhouse at Instituto Superior de Agronomia of the University of Lisbon (ISA/UL), in order to obtain developed and mature leaves needed to further evaluate their antioxidant capacity. All the species were cultivated in pots with fertile and well-drained soils with the optimal temperature for germination varying between 25 °C to 27 °C. All seeds germinated after 10-20 days. A total of 15 accessions of beans and leaves were analysed, namely of: *Cajanus cajan* (n=2), *Lablab purpureus* (n=6), *Phaseolus lunatus* (n=2), *Phaseolus vulgaris* (n=2) and *Vigna unguiculata* (n=3). Their origin and location in Santiago are indicated in Figure 2.2.

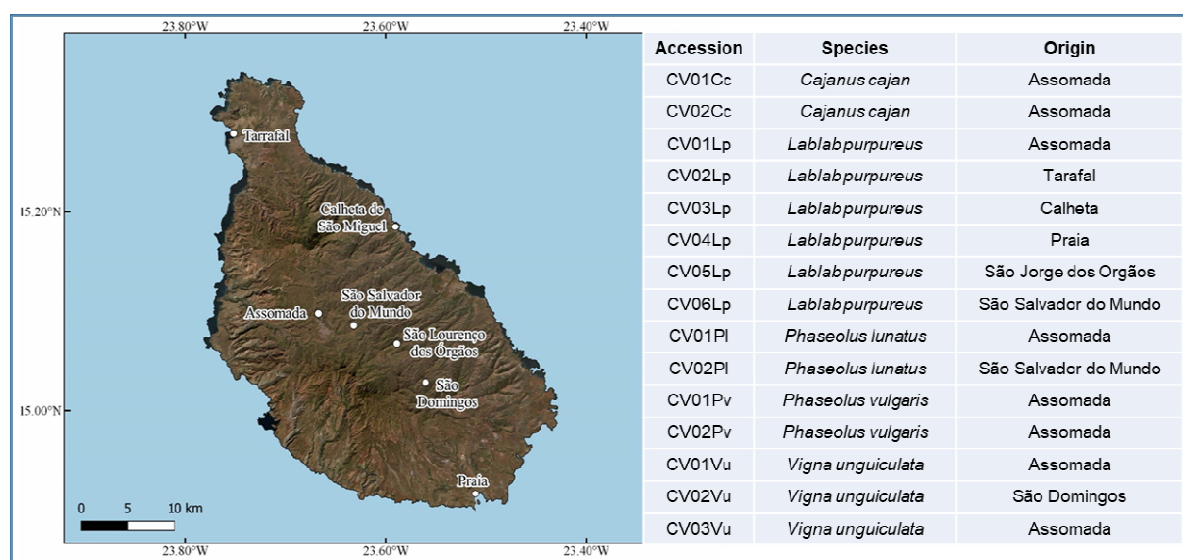


Figure 2.2: Santiago Island. List of the 15 Cabo Verde bean accessions used for this study and their origin/market location.

2.3. Chemical composition and antioxidant analyses

2.3.1. Reagents

Acetone, ascorbic acid, gallic acid, sodium hydroxide, potassium hydroxide, potassium sulphate, selenium metal powder, sodium carbonate and standard solutions for ICP (boron, calcium, copper, iron, magnesium, manganese, phosphorus, potassium, sulphur and zinc) were from Panreac (Barcelona, Spain), 2,2-diphenyl-1-picrylhydrazyl (DPPH) was from Sigma-Aldrich (St. Louis, MO, USA) and boric acid were from Chem-lab (Zedelgem, Belgium), ethanol absolute and sulphuric acid were from Riedel-de Haën (Seelze, Germany),

hydrochloric acid, nitric acid and Folin-Ciocalteu reagent were from Merck (Darmstadt, Germany). All reagents used in the analytical procedures were of analytical reagent grade. All water used was purified using a Milli-Q water system (Millipore, Bedford, MA, USA).

2.3.2. Sample preparation

Whole beans (100 g seeds of each bean accession) were ground into flour using a stainless-steel grinder (Kunft coffee mill) and analysed for moisture, ash, protein, fibre and mineral contents.

Leaves from the different species were collected and allowed to dry for two weeks, in the dark, at room temperature. Then, dry leaves were ground into powder in a porcelain mortar with pestle. Grounded dry leaves (5 g) were extracted with 100 mL of ethanol (70% v/v), for 24 h, in the dark, at room temperature and under stirring. The resulting extracts were filtered (Whatman, n°1) and stored at – 50 °C until future analyses.

2.3.3. Moisture and ash

Moisture and ash contents were determined according to the standard gravimetric method (AOAC, 2005). Briefly, moisture content was measured by drying the sample at 103 ± 2 °C (Laboratory heater, Memmert UL500, Schwabach, Germany) for 2 h and repeated until constant weight, while ash content was determined by incinerating the sample for 6 h at 550 ± 25 °C (Muffle furnace, Heraeus Hanau MR170E, Hanau, Germany). The analyses were performed in duplicate and the results expressed in g per 100 g wet weight.

2.3.4. Crude fibre

The crude fibre determination was performed by Weende's method (AOAC, 2005). Briefly, test portions of each sample were submitted to acid hydrolysis with 150 mL of H₂SO₄ (0.128 M), at boiling temperature and for 30 minutes. Then, the mixtures were filtered through a glass Gooch crucible (porosity P2), under vacuum, washed with water and submitted to basic hydrolysis with 150 mL of KOH (0.223 M), at boiling temperature and for 30 minutes. Subsequently, the mixtures were once more filtered through a glass Gooch crucible (porosity P2), under vacuum, washed with water and finally with acetone. The crucibles with the fibre

were dried at 103 ± 2 °C (Laboratory heater, Memmert UL500, Schawabach, Germany) and weighted after cooling in a desiccator. Then, the sample was incinerated (3 h at 550 ± 25 °C, muffle furnace, Heraeus Hanau MR170E, Hanau, Germany), cooled in a desiccator, and reweighted. The total crude fibre content was expressed in g per 100 g wet weight.

2.3.5. Protein

The protein content was determined by the Kjeldahl method (Lynch & Barbano, 1999). Briefly, test portions of each sample were digested with concentrated sulphuric acid, in the presence of potassium sulphate and a low concentration of selenium catalyst, at 360 °C (Digestion System Tecator 2006, Höganäs, Sweden). During the digestion, nitrogen is released and retained as ammonium sulphate. After cooling to room temperature, ammonia was released from the acid digest by raising the pH with the addition of sodium hydroxide (6 M). Then, ammonia was distilled (Tecator Distilling Unit 1002, Höganäs, Sweden), collected in a boric acid solution, and titrated with a standardized sulphuric acid solution (0.02 N). Protein content was calculated using a conversion factor of 6.25 (FAO, 1973). The analyses were performed in duplicate and the results were expressed in g per 100 g wet weight.

2.3.6. Minerals

Minerals (B, Ca, Cu, Fe, K, Mg, Mn, P, S, Zn) in *Cajanus cajan*, *Lablab purpureus*, *Phaseolus vulgaris*, *Phaseolus lunatus* and *Vigna unguiculata* seeds were quantified by inductively coupled plasma optical emission spectrometry. Test portions of each sample were weighed and submitted to a digestion process with a mixture of nitric acid and hydrochloric acid (1:3, v/v) at 105 °C during 90 min and analysed by Inductively coupled plasma - optical emission spectrometry (ICP-OES) using a Thermo Scientific iCAP 7000 Series ICP-OES spectrometer (Thermo Scientific, Cambridge, UK). Procedural blanks were prepared using the same analytical procedure and reagents. Calibration curves of, at least, five different concentrations were used to quantify each element. The analyses were performed in triplicate and the results expressed in mg per kg wet weight.

2.3.7. Total phenolic content

Total phenolic compounds were determined according to Loebler et al. (2020). Briefly, water (6.0 mL), leaf ethanolic extract (100 μ L) and undiluted Folin-Ciocalteu reagent (500 μ L) were mixed in a 10.0 volumetric flask. After 1 min, 1500 μ L of 20% (w/v) Na_2CO_3 was added and the volume was made up to 10.0 mL with H_2O . After 2 h incubation at room temperature and in the dark, the absorbance was measured at 765 nm (SPEKOL 1500, Analytik Jena, Germany) and compared to a gallic acid calibration curve. The analyses were performed in triplicate and the results were expressed in mg of gallic acid equivalents (GAE) per g of dry leaves.

2.3.8. DPPH radical scavenging capacity

The antioxidant capacity was determined according to the methodology described by Lima et al. (2019). Briefly, a 500 μ L aliquot of diluted leaf ethanolic extract was added to 3 mL of daily prepared DPPH solution (24 mg/L in ethanol). After 30 min incubation at room temperature and in the dark, the absorbance was measured at 517 nm (SPEKOL 1500, Analytik Jena, Germany) and compared to an ascorbic acid calibration curve. The scavenging activity was measured as the decrease in absorbance of the samples versus DPPH standard solution. The analyses were performed in triplicate and the results were expressed in mg of ascorbic acid equivalents (AAE) per g of dry leaves.

2.4. Physical analysis

2.4.1. Color measurements

The color of the beans was determined in triplicate, by measurement of CIE- $L^*a^*b^*$ coordinates ($L^* = 0$ (black) to $L^* = 100$ (white), $-a^*$ (greenness) to $+a^*$ (redness), and $-b^*$ (blueness) to $+b^*$ (yellowness)), on the surface of 10 beans using a portable Konica Minolta CR-300 Chroma Meters colorimeter (Minolta Co. Ltd., Osaka, Japan), previously calibrated in a white leaf tile pattern. The coordinates values were used to calculate the CIEL $^*C^*h^\circ$ color space, where C^* correspond to Chromaticity (color saturation) whilst h° presents the hue angle as a shade according to the angle in the 360° color wheel, with a red-purple at 0°, yellow shade at 90°, gray-green shade are 180° and blue shade at 270°, counterclockwise (McGuire,

1992). The values of Chromaticity (C^*) and the hue angle (h°) were obtained based on Equation (1) and Equation (2).

$$\text{Chromaticity } (C^*) = [(a^*)^2 + (b^*)^2]^{1/2} \quad (1)$$

$$\text{hue angle } (h^\circ) = \tan^{-1} (b^*/a^*), \text{ when } a^* > 0 \text{ and } b^* < 0 \quad (2)$$

2.4.2. Morphometric measurements

The length and width of 10 beans from each species were measured using a digital caliper with 0.01 mm accuracy. Seed length was measured from the base to the tip portion while the seed width was measured from the hilum to the opposite side. The mean values were recorded in millimeters.

2.5. Agronomic data collection

The information on economic and agricultural profiles of each bean species under study was also investigated during the field surveys made between 2018 and 2019 at the main trade markets of Santiago Island. This information was complemented with data concerning the agro-economical characterization of Cabo Verde Islands, which was retrieved from: (1) agricultural data from the Ministry of Agriculture and Environment (MAA, 2016 and 2017); (2) the National Institute of Statistics (INE, 2018a and 2018b); and (3) the Annual Report of Cabo Verde (ANSA, 2005 and 2011).

2.6. Statistical analyses

All data measurements are presented as mean values. Univariate analysis (UA) was performed to compare the chemical and nutritional traits among the bean species. Before running the UA, normality and homogeneity of variances were tested; as data did not follow normal distributions and the variances were not homogeneous, the test of means was carried out using Kruskal Wallis test for all variables ($\alpha=0.05$). After standardization (mean = 0, and standard deviation = 1) of chemical data, a multivariate analysis by principal component analysis (PCA), based on the correlation matrix, was performed and the eigenvectors and eigenvalues

projected and visualized with the `ggplot` function of the `ggplot2` package (Wickham, 2016). All analyses were carried out in the RStudio program version 1.1.456 (R Core Team, 2020).

CHAPTER III

3. Results and Discussion

3.1. Results

3.1.1. Characterization of markets and trade bean species

All the 27 sellers surveyed (7 in the market of Assomada, 8 in Praia, 3 in Calheta, 4 in Tarrafal, 3 in São Domingos, and 2 in São Lourenço dos Orgãos) were women, 22 were between 25 and 59 years old and 5 were over 59 years old. Most of them have only completed the basic education, and another part is illiterate. Most consider themselves to be the head of the household.

Five Leguminosae species are traded in the different markets of the Santiago island: *Cajanus cajan*, *Lablab purpureus*, *Phaseolus lunatus*, *Phaseolus vulgaris* e *Vigna unguiculata*. Regarding beans prices, there was a difference between species and seasons of demand. Thus, the price varies between 150 - 450\$, being the *Phaseolus lunatus* and *Cajanus cajan* the most expensive. Prices varied from market to market, due to factors such as the distance from the producer or the demand. Generally, the greater the demand, the higher the price.

Some sellers sell their own cultivated products directly on the market and there are others that buy directly from the farmers and resell on the market. It should be emphasized that most of the resellers on the Praia and Assomada market look for the products on the interior of the island, where there are more quantity and diversity of cultivated beans.

In the "azáguas" times, the demand for Leguminosae in the markets increases, since they are often used to make the cultivation, which takes place in the month of July to August. In other seasons the demand is more directed to food, where it is often used in the preparation of meals in wedding ceremonies, baptism and among others purposes. Sellers have knowledge of the importance of Leguminosae, but they also showed a little bit of disappointment, since they felt that many consumers don't have that knowledge.

According to the information collected, on fair days (each market has its fixed days) there is a larger crowd of people in the morning, in order to find fresher products. The demand for

Leguminosae plants is more for national people, while tourists visit more the artisan and local products areas. Besides beans, there is also a high demand for corn, one of the typical Cabo-Verdean foods.

3.1.2. Diversity of food legume species

Our research showed that 15 Leguminosae species were recognized as food plants in Cabo Verde and their native distribution, habit and distribution are presented in Table 3.1. Most of these species are non-native (73%, 11 species) and only four native species (27%) were accounted for. Also, 93% (14 species) of all species are cultivated, and *Zornia glochidiata* is the only one that is not so. Only 21% of the cultivated species (three species) are native to Cabo Verde. As previously mentioned, all 15 species are used for food, however, seven species (47%) are also described as medicinal, and eight species (53%) as forage and medicinal.

Considering the worldwide native distribution of each species (Table 3.1), four main groups were identified: Neotropical species (33%, five species) (e.g., *Arachis hypogaea*, *Phaseolus lunatus* and *Phaseolus vulgaris*); Oriental species (26.5%, four species) (e.g., *Cajanus cajan*, *Cassia fistula* and *Sesbania grandiflora*); Afrotropical species (26.5%, four species) (e.g., *Tamarindus indica*, *Lablab purpureus* and *Vigna unguiculata*); and Palearctic species (7%, one species: *Ceratonia siliqua*). Only one species, *Mucuna pruriens*, is distributed across two distinct biogeographical regions. The majority of the non-native food legume species used in Cabo Verde is from Neotropical and Oriental regions.

Morphologically, these 15 food legume species display a great diversity of habit. More than half (54%) are herbaceous, annual or biennial, 33% are trees, and only 13% correspond to shrubs (Table 3.1). The majority of the studied food legumes (87%) are commonly found in Santiago (13 species), followed by Santo Antão (73%, 11 species) and Fogo (73%, 11 species); Boavista hosts the lowest number of species (13%, two species) (Figure 3.1; Table 3.1).

Table 3.1: Food legume species in Cabo Verde: scientific name, common name, status in Cabo Verde, important uses in Cabo Verde, native distribution, and global conservation status.

Species	Common names	Status ^a	Important uses	Habit	Native distribution	Distribution in Cabo Verde ^b
<i>Arachis hypogaea</i> L.	Groundnut, Peanut	Ic	Food, Forage, Medicinal	Annual herb	Neotropical	A, T
<i>Cajanus cajan</i> (L.) Millsp.	Pigeon Pea	Ic	Food, Forage, Medicinal	Shrub	Oriental	A, N, B, T, F, Br
<i>Canavalia ensiformis</i> L.	Jack bean	Ic	Food, Medicinal	Annual herb	Oriental	A, M, T, Br
<i>Cassia fistula</i> L.	Golden shower	Ic	Food, Medicinal	Tree	Oriental	A, T, F
<i>Ceratonia siliqua</i> L.	Carob tree	Ic	Food, Forage, Medicinal	Tree	Paelearctic	A, N, T, F, Br
<i>Lablab purpureus</i> (L.) Sweet	Hyacinth Bean	Nc	Food, Forage, Medicinal	Annual herb	Afrotropical	A, V, N, S, B, T, F, Br
<i>Mucuna pruriens</i> (L.) DC.	Velvet bean	Nc	Food, Forage, Medicinal	Annual herb	Afrotropical-Oriental	A, T
<i>Phaseolus lunatus</i> L.	Lima Bean	Ic	Food, Medicinal	Annual or biennial herb	Neotropical	A, T, F, Br
<i>Phaseolus vulgaris</i> L.	Common bean, Kidney bean	Ic	Food, Medicinal	Annual or biennial herb	Neotropical	A, T, F, Br
<i>Pithecellobium dulce</i> (Roxb.) Benth.	Manila tamarind	Ic	Food, Medicinal	Tree	Neotropical	F, Br
<i>Samanea saman</i> (Jacq.) Merr.	Monkeypod	Ic	Food, Forage, Medicinal	Tree	Neotropical	Br
<i>Sesbania grandiflora</i> (L.) Pers.	Agati sesbania	Ic	Food, Medicinal	Shrub	Oriental	T, F
<i>Tamarindus indica</i> L.	Tamarind	Ic	Food, Forage, Medicinal	Tree	Afrotropical	A, V, N, S, M, T, F, Br
<i>Vigna unguiculata</i> (L.) Walp.	Cowpea	Nc	Food, Forage, Medicinal	Annual herb	Afrotropical	A, V, N, S, M, T, F, Br
<i>Zornia glochidiata</i> Reichb. ex DC.	Herbe mouton	N	Food, Medicinal	Annual herb	Afrotropical	A, T, F

^aI, Introduced; N, Native; c, cultivated; ^bDistribution in Cabo Verde: Islands: A, Santo Antão; V, São Vicente; N, São Nicolau; S, Sal; B, Boavista; M, Maio; T, Santiago; F, Fogo; Br, Brava.

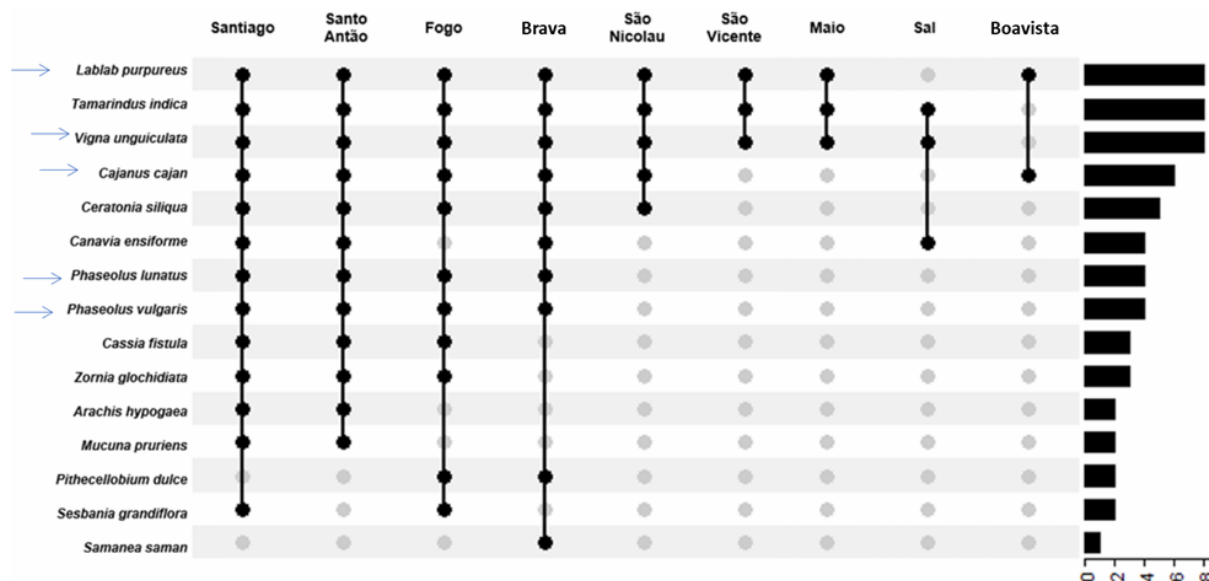


Figure 3.1: Distribution of the food legume species in Cabo Verde. UpSet diagram showing the presence and number of species per island. The lines linking the dots represent species that occur in two or more islands.

3.1.3. Chemical composition

Ash, fibre, moisture and protein contents (%) of the five most cultivated and traded legume species in Cabo Verde were compared through boxplot analysis as shown in Figure 3.2. Overall, the contents did not differ considerably among these species ($p < 0.05$). However, mean ash contents of *Vigna unguiculata* (3.2 ± 0.3 g/100 g wet weight) were the lowest whereas *Phaseolus vulgaris* (4.1 ± 0.0 g/100 g wet weight) showed the highest ones, *Lablab purpureus* (7.8 ± 1.0 g/100 g wet weight) and *Cajanus cajan* (6.3 ± 0.1 g/100 g wet weight) were the richest species in fibre content and *Phaseolus lunatus* and *Phaseolus vulgaris* the poorest (3.7 ± 1.0 g/100 g wet weight and 3.7 ± 0.4 g/100 g wet weight, respectively). The average moisture contents ranged from 10.7 ± 0.2 g/100 g (*Phaseolus vulgaris*) to 12.4 ± 0.9 g/100 g (*Phaseolus lunatus*). The highest contents in protein were measured in *Lablab purpureus* (23.3 ± 0.6 g/100 g wet weight), followed by *Phaseolus vulgaris* (23.2 ± 0.4 g/100 g wet weight), *Vigna unguiculata* (23.0 ± 1.4 g/100 g wet weight), *Cajanus cajan* (22.0 ± 2.0 g/100 g wet weight) and *Phaseolus lunatus* (19.0 ± 1.8 g/100 g wet weight).

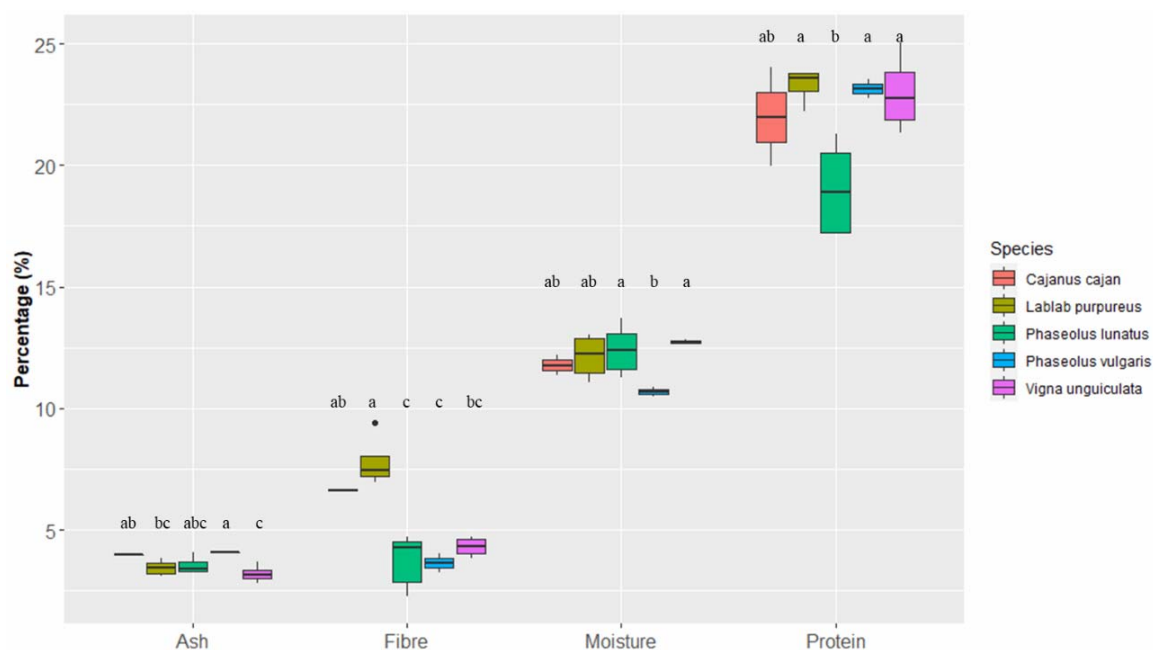


Figure 3.2: Intraspecific and interspecific variation of ash, fibre, moisture and protein contents in the beans of the five most cultivated and traded legume species of Cabo Verde. The box represents the 25th, 50th (median) and 75th percentiles, while whiskers represent the 10th and 90th percentiles with minimum and maximum observations. The black dots represent the outliers. Species sharing one or more letters for each trait are not statistically different ($p < 0.05$).

The highest leaf phenolic contents (Table 3.2) were found in *Cajanus cajan* (4.55 ± 0.16 mg GAE/mg dry weight) and *Lablab purpureus* (4.13 ± 0.28 mg AGE/mg dry weight). On the other hand, the lowest contents were 3.15 ± 0.48 mg GAE/mg dry weight (*Phaseolus lunatus*), 3.04 ± 0.15 mg GAE/mg dry weight (*Vigna unguiculata*) and 3.02 ± 0.08 mg GAE/mg dry weight (*Phaseolus vulgaris*). Consistent with the phenolic contents, the antioxidant capacities of *Cajanus cajan* and *Lablab purpureus* were the highest (Table 3.2), respectively, 2.35 ± 0.08 mg AAE/mg dry weight and 2.49 ± 0.11 mg AAE/mg dry weight. Accordingly, *Phaseolus lunatus* (1.75 ± 0.44 mg AAE/ mg dry weight), *Vigna unguiculata* (1.84 ± 0.11 mg AAE/ mg dry weight), and *Phaseolus vulgaris* (2.22 ± 0.05 mg AAE/ mg dry weight) presented the lowest antioxidant capacities.

Table 3.2: Mean values, standard deviations (SD) and homogeneous groups¹ of the antioxidant capacities of the five most cultivated and traded bean species of Cabo Verde.

	<i>Cajanus cajan</i>	<i>Lablab purpureus</i>	<i>Phaseolus lunatus</i>	<i>Phaseolus vulgaris</i>	<i>Vigna unguiculata</i>
Total Phenolic Content (mg GAE/mg dry weight) ²	4.55 ± 0.16^a	4.13 ± 0.28^a	3.15 ± 0.48^b	3.02 ± 0.08^b	3.04 ± 0.15^b
DPPH radical scavenging capacity (mg AAE/ mg dry weight) ³	2.35 ± 0.08^a	2.49 ± 0.11^a	1.75 ± 0.44^b	2.22 ± 0.05^{ab}	1.84 ± 0.11^b

¹Homogeneous groups: species sharing one or more letters for each variable are not statistically different ($p < 0.05$). ²GAE, Gallic acid equivalents. ³AAE, Ascorbic acid equivalents.

Table 3.3 presents the mineral contents of the five most cultivated and traded legume species of Cabo Verde: *Cajanus cajan*, *Lablab purpureus*, *Phaseolus lunatus*, *Phaseolus vulgaris* and *Vigna unguiculata*. The B contents ranged from 5.8 ± 0.6 mg/kg wet weight (*Lablab purpureus*) to 9.2 ± 1.8 mg/kg wet weight (*Vigna unguiculata*). The average contents of Ca ranged from 581.9 ± 97.8 (*Lablab purpureus*) to 1418.5 ± 63.3 mg/kg wet weight (*Phaseolus vulgaris*). The Cu contents ranged between 4.8 ± 0.7 (*Vigna unguiculata*) and 9.4 ± 2.4 mg/kg wet weight (*Cajanus cajan*). Fe values varied between 39.6 ± 1.3 mg/kg wet weight (*Cajanus cajan*) and 86.5 ± 21.0 mg/kg wet weight (*Phaseolus lunatus*). K contents ranged between 7607.5 ± 599.1 mg/kg wet weight (*Phaseolus lunatus*) and 11704.7 ± 201.1 (*Phaseolus vulgaris*). On average, *Cajanus cajan* has the lowest Mg content (1229.2 ± 35.5 mg/kg wet weight) and *Vigna unguiculata* the highest (1899.3 ± 123.4 mg/kg wet weight). *Cajanus cajan* showed the lowest Mn content (15.7 ± 0.3 mg/kg wet weight) and *Lablab purpureus* the highest (26.4 ± 1.5 mg/kg wet weight). The lowest P content was 3662.4 ± 335.2 mg/kg wet weight (*Cajanus cajan*) and the highest was 4370.3 ± 181.1 mg/kg wet weight (*Phaseolus vulgaris*). The S content of *Phaseolus lunatus* was the lowest (1483.4 ± 326.1 mg/kg wet weight) while *Vigna unguiculata* exhibited the highest (1937.2 ± 46.0 mg/kg wet weight). Finally, *Phaseolus vulgaris* showed a Zn content of 21.7 ± 1.8 mg/kg wet weight and *Vigna unguiculata* a mean value of 27.2 ± 3.5 mg/kg wet weight, respectively the highest and lowest values.

Table 3.3: Mean values (mg/kg wet weight), standard deviations (SD) and homogeneous groups¹ for the mineral contents in the beans of the five most cultivated and traded legume species of Cabo Verde.

Minerals	<i>Cajanus cajan</i>	<i>Lablab purpureus</i>	<i>Phaseolus lunatus</i>	<i>Phaseolus vulgaris</i>	<i>Vigna unguiculata</i>
B	6.2 ± 0.3^{bc}	5.8 ± 0.6^c	6.1 ± 1.1^{bc}	7.0 ± 1.2^b	9.2 ± 1.8^a
Ca	1144.4 ± 67.0^a	581.9 ± 97.8^c	1332.0 ± 438.5^a	1418.5 ± 63.3^a	818.1 ± 72.2^b
Cu	9.4 ± 2.4^a	6.8 ± 2.5^{ab}	6.0 ± 1.4^{bc}	6.6 ± 0.6^{ab}	4.8 ± 0.7^c
Fe	39.6 ± 1.3^c	54.4 ± 3.1^b	86.5 ± 21.0^a	76.0 ± 3.6^a	54.0 ± 2.0^b
K	9148.6 ± 1466.8^{bc}	8255.3 ± 1314.5^{cd}	7607.5 ± 599.1^d	11704.7 ± 201.1^a	10149.0 ± 282.6^b
Mg	1229.2 ± 35.5^b	1820.7 ± 73.4^a	1726.0 ± 230.6^a	1798.4 ± 43.7^a	1899.3 ± 123.4^a
Mn	15.7 ± 0.3^c	26.4 ± 1.5^a	18.9 ± 0.9^b	16.2 ± 1.7^c	17.1 ± 1.7^{bc}
P	3662.4 ± 335.2^b	4004.5 ± 151.0^b	3782.8 ± 267.5^b	4370.3 ± 181.1^a	4167.4 ± 728.1^b
S	1555.3 ± 61.0^c	1689.9 ± 168.7^b	1483.4 ± 326.1^{bc}	1859.8 ± 54.0^a	1937.2 ± 46.0^a
Zn	25.6 ± 0.5^a	26.0 ± 5.0^a	22.9 ± 1.3^b	21.7 ± 1.8^b	27.2 ± 3.5^a

¹Homogeneous groups: species sharing one or more letters for each mineral are not statistically different ($p < 0.05$).

In order to assess the patterns of variations in the mineral contents of the five Cabo Verde bean species (Table 3.3), a Principal Components Analysis (PCA) was performed. The first four principal components (PCs) accounted for 77.40% of the variability amongst the five species (Supplementary Table S1). The first PC (PC1) accounted for 29.36% of the total variation with B, K and P presenting negative coefficients. PC2 accounted for additional 20.76% of the total variation, with Ca and Mn being the most important contributors, the first with a negative coefficient and the latter with a positive one. PC3 accounted for further 16.58% of the variability and showed Cu as a strong negative contributor and Mg as a positive one. PC4 accounted for 10.70% of the variability, describing the patterns of variation of Fe and Zn with positive coefficients, and S with negative one. The differentiation patterns between the five species for PC1 and PC2 are presented in Figure 3.3, showing a large mineral diversity that slightly distinguishes the species. *Lablab purpureus* showed positive values of both components while *Phaseolus vulgaris* showed negative values. *Cajanus cajan* displayed positive values for PC1 and negative values for PC2, contrasting with *Vigna unguiculata* which showed positive values for PC2 and negative values for PC1. Finally, *Phaseolus lunatus* presented a broad variety of mineral contents with both negative and positive values of PC1 and PC2.

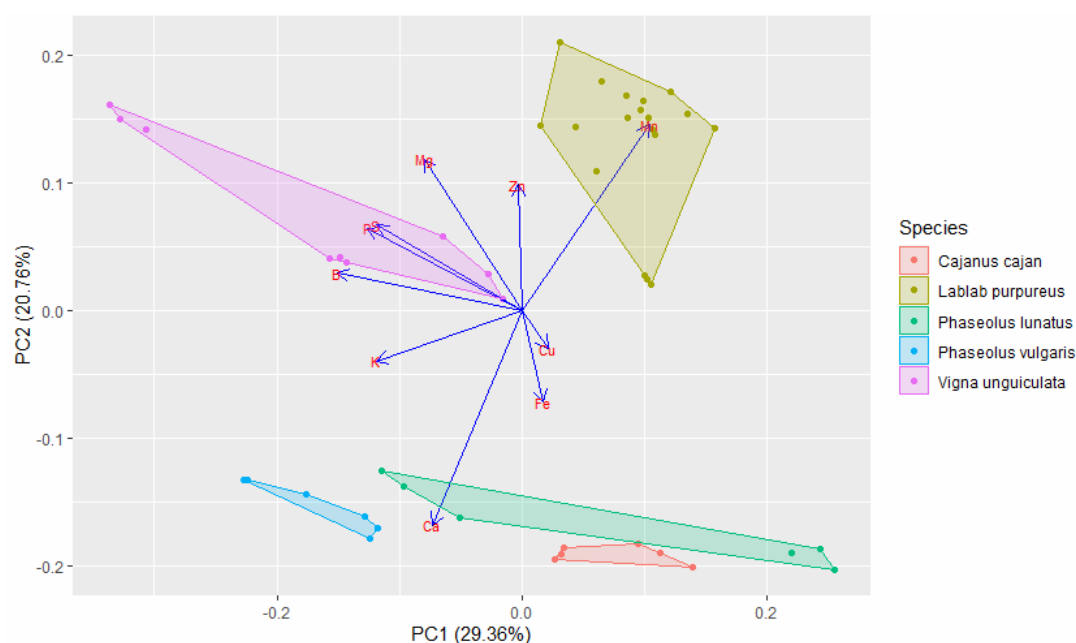


Figure 3.3: Representation of the two first components (PC1 and PC2) resulting from the principal component analysis, which explain 50.12% of the diversity of mineral contents in beans of the five most cultivated and traded legume species of Cabo Verde in a space de defined by vectors and own values. The arrow lengths show differences in variance explained relative to each other.

3.1.4. Physical analysis

Table 3.4 shows the mean values and standard deviation, for the physical characteristics of the seeds of the five most cultivated and traded beans species of Cabo Verde, specifically two morphometric measurements (length and width) and five colorimetric measurements (L^* , a^* , b^* , C^* and h). Regarding length and width values, *Phaseolus lunatus* presented the higher values (17.4 ± 1.7 and 13.6 ± 0.7 mm, respectively) and *Cajanus cajan* the lowers (7.0 ± 0.1 and 6.3 ± 0.3 mm, respectively). The luminosity (L^* value) varied from 35.8 ± 1.5 (*Phaseolus vulgaris*) to 66.8 ± 10.7 (*Phaseolus lunatus*). Values of a^* ranged from 3.7 ± 2.6 (*Lablab purpureus*) to 15.6 ± 1.5 (*Phaseolus vulgaris*), while values of b^* ranged from 2.8 ± 1.0 (*Phaseolus vulgaris*) to 17.0 ± 7.0 (*Lablab purpureus*). The chromaticity (C^*) value varied from 12.3 ± 2.5 (*Cajanus cajan*) to 18.7 ± 4.9 (*lablab purpureus*). For angle of Hue (h), on average, *Phaseolus vulgaris* demostarted the lowest values (0.2 ± 0.1) while the *Phaseolus lunatus* had the highest values (1.3 ± 0.2).

Table 3.4: Mean values, standard deviation (SD) and homogeneous groups¹ for physical features of five most cultivated and traded bean species of Cabo Verde.

Features	<i>Cajanus cajan</i>	<i>Lablab purpureus</i>	<i>Phaseolus lunatus</i>	<i>Phaseolus vulgaris</i>	<i>Vigna unguiculata</i>
Length	7.0 ± 0.1^d	12.2 ± 0.4^c	17.4 ± 1.7^a	15.4 ± 1.1^b	7.4 ± 0.3^d
Width	6.3 ± 0.3^{cd}	9.9 ± 0.3^b	13.6 ± 0.7^a	6.8 ± 0.6^c	6.2 ± 0.4^d
L^*	48.4 ± 4.0^{bc}	60.3 ± 11.8^a	66.8 ± 10.7^a	35.8 ± 1.5^c	53.7 ± 9.8^b
a^*	4.9 ± 1.3^b	3.7 ± 2.6^c	5.1 ± 3.6^{bc}	15.6 ± 1.5^a	6.4 ± 4.0^b
b^*	11.3 ± 2.2^c	17.0 ± 7.0^a	16.0 ± 4.2^{ab}	2.8 ± 1.0^c	16.0 ± 2.6^b
C^*	12.3 ± 2.5^c	18.7 ± 4.9^a	17.1 ± 4.4^{ab}	15.9 ± 1.7^{bc}	17.6 ± 3.2^{ab}
h	1.2 ± 0.1^b	1.2 ± 0.4^{ab}	1.3 ± 0.2^{ab}	0.2 ± 0.1^c	1.2 ± 0.2^b

¹Homogeneous groups: species sharing one or more letters for each mineral are not statistically different ($p < 0.05$).

3.1.5. Agronomic analysis

Figure 3.4 show that the evolution of the bean cropland area over the last decade followed a trend similar to that of rainfed crops. The bean cropland in Cabo Verde represents about 50% of the total extant rainfed cropland. In 2006, 2010, 2013 and 2015 the overall cultivated area decreased. Over the studied period, the maximum cultivated area was reached in 2008, corresponding to 66 411 ha in the whole and to 34 385 ha in bean cropland.

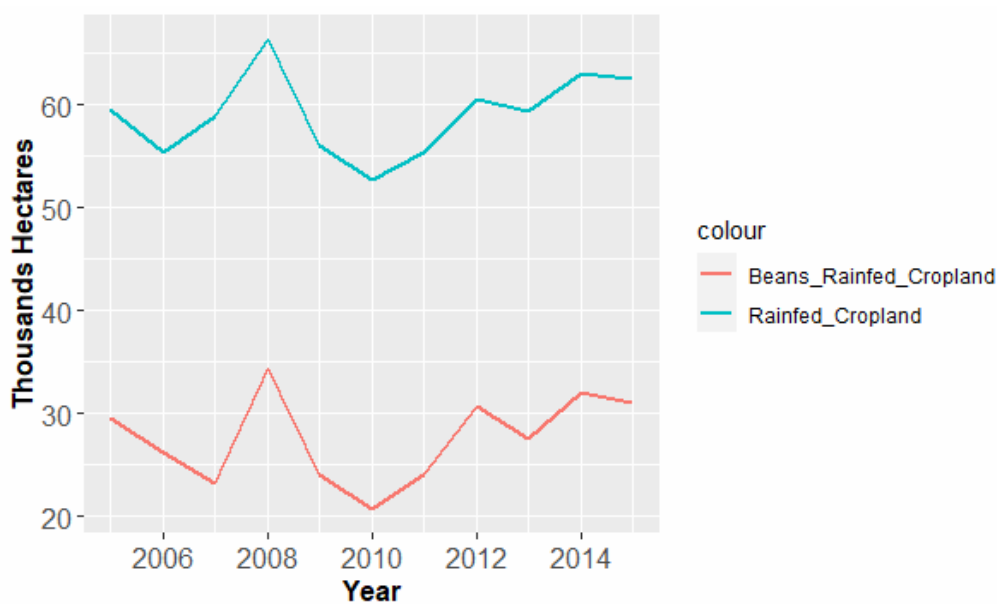


Figure 3.4: Areas of extant rainfed cropland and beans rainfed cropland in Cabo Verde, from 2005 to 2015 (INE, 2018b; MAA, 2016).

According to Figure 3.5, the annual production of beans as traditional rainfed crops severely fluctuated from 2013 to 2017, decreasing from 5943 to 700 tons in 2014 and increasing to 5199 tons in 2015. The lowest bean production happened in 2017, and represented a drastic reduction of 99.8% when compared with 2016, while in 2012 the maximum was observed (5950 tons), over the studied period. Overall, a negative linear trend of beans production was observed from 2006 to 2017.

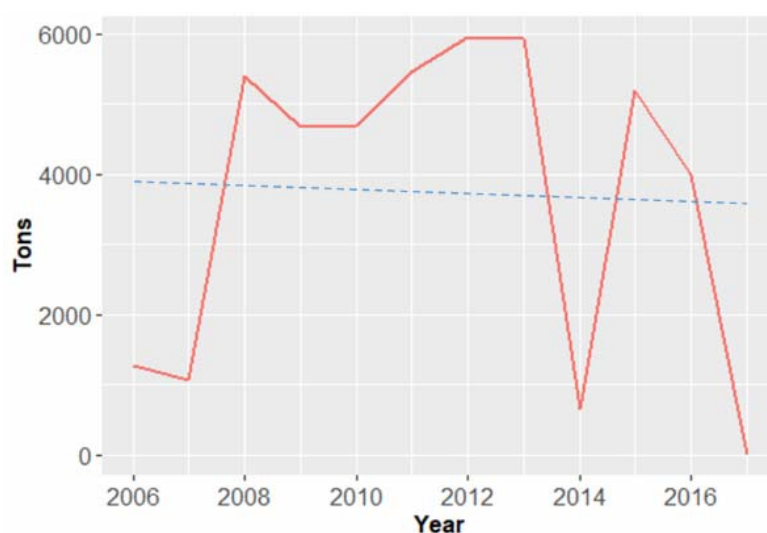


Figure 3.5: Annual bean production (in tons) of the five most cultivated and traded legume species in Cabo Verde, from 2006 to 2017 (INE, 2018a; MAA, 2016).

3.2. Discussion

3.2.1. Food legume species in Cabo Verde

Legumes supply nutrients and less expensive, non-animal proteins to meet the needs of people, particularly of Low- and Middle- Income Countries, and surpluses can be sold to generate family income (Schreinemachers et al., 2014). The present study allowed us to identify five pulses (*Cajanus cajan*, *Lablab purpureus*, *Phaseolus lunatus*, *Phaseolus vulgaris* and *Vigna unguiculata*) that are widely traded in Santiago markets, but also to identify the Leguminosae species used as food in Cabo Verde. Fifteen legume species are used as food, with a significant share described as medicinal (47%) and as forage (53%). All of those species are cultivated, except for *Zornia glochidiata*, which is common in Sudano-sahelian pastures (Akpo et al., 2002). There is a predominance of species from Tropical regions, the places of origin and domestication of a large number of the species cultivated in the archipelago, like beans (*Canavalia ensiformis*, *Lablab purpureus*, *Phaseolus lunatus*), peanuts (*Arachis hypogaea*) or manioc (*Manihot esculenta*: family Euphorbiaceae) (Monteiro et al. 2020). Presently, *Cajanus cajan*, *Vigna unguiculata*, *Lablab purpureus*, *Phaseolus lunatus* and *Phaseolus vulgaris* continue to have significant importance at both food and medicinal levels, as reported in previous studies (Teixeira & Barbosa, 1958; Romeiras et al., 2011).

3.2.2. Chemical composition

Legumes are known to significantly contribute to the supply of bioactive compounds to the body due to their antioxidant activity, attributed to phenolic compounds, and are also rich sources of proteins, dietary fibres, and micronutrients. Legumes have also acquired a significant importance in African traditional medicine (Catarino et al., 2019; Akinola et al., 2020). Our results revealed that *Lablab purpureus*, *Phaseolus vulgaris* and *Vigna unguiculata* have higher protein contents (~23%), with the former containing also high levels of phenolic contents and antioxidant capacities in its leaves, together with *Cajanus cajan*. Despite the high antioxidant and phenolic contents of legume leaves, they are currently used to feed animals during fodder shortages in the dry season. Seeds, as part of daily diet, are the most consumed plant parts in Cabo Verde (Silva, 2005). Neglecting legume leaves as food has been more frequent in Cabo Verde than in other West African countries, where they are commonly

cooked in stews (Sprent et al., 2010). In a country where food shortage and malnutrition still prevails, such as Cabo Verde, the promotion of leaves in the diet should be considered due to its potential high health benefits as anticarcinogenic, anti-inflammatory, antioxidant, and anti-microbial (Bittenbender et al., 1984).

Results revealed that ash contents ranged from 3.2% (*Vigna unguiculata*) to 4.1% (*Phaseolus vulgaris*), indicating that they are good sources of minerals (Barampama & Simard, 1993; Gondwe et al., 2019). Moreover, all the studied species revealed an average moisture content ranging from 10.7 ± 0.2 g/100 g in *Phaseolus vulgaris* to 12.4 ± 0.9 g/100 g in *Phaseolus lunatus* and we concluded that it is possible to store all these beans with quality, in agreement with other studies (Ngwenyama et al., 2020). Amarteifio et al. (2002) recorded fibre contents of *Cajanus cajan* cultivated in Botswana ranging from 9.8 to 13.0 g/100g, higher than those we obtained (6.3 g/100g). This difference can be explained by the different provenances of the samples, taking the particular environmental aspects of each region into account.






The presence of iron and zinc in the screened beans species is vital, as these micronutrients are responsible for essential body functions, and a deficiency in these minerals can lead to severe medical conditions (Clemens, 2014). Iron is needed for the transfer of oxygen to body tissues and organs; it is the most common nutrient deficiency, affecting over 2 billion people worldwide, and a major public health burden concerning African children (Muriuki et al., 2020). Zinc plays an essential role in body metabolism and it prevents illnesses by supporting the immune system (Chasapis et al., 2012). The zinc and iron contents show significant differences between beans species, with *Phaseolus vulgaris* and *Phaseolus lunatus* being the iron-richest species, but *Phaseolus vulgaris* having lower zinc content. These results highlight a species-specific profile of zinc and iron contents, which does not significantly affect the nutritional value of each bean species. A study conducted in Cabo Verde by Semedo et al. (2014) revealed a prevalence of anaemia of 51.8%, particularly high in children below 24 months of age living under poor household conditions, thus highlighting anaemia as a public-health concern in the country. In addition to the traditional use of beans as a multipurpose crop, the increase of its consumption at household level could be foreseen to overcome some dietary deficiencies (iron).

3.2.3. Physical Characterization

One of the relevant information for beans which is associated with the clarity of the grains is the value of L. There was a difference in brightness between the five species of beans.

Phaseolus lunatus and *Lablab purpureus* presented the highest value of L^* , which means it has the lightest color of all beans. On the other hand, *Phaseolus Vulgaris* presented the lowest value of L^* , which means it has the darker color. According to Burr et al. (1968), the color is a characteristic of each crop. Changes in color may be due to several factors that influence the initial color of the beans. Examples of these factors are the climate condition, the soil type and the post-harvest condition. Bressani (1993) showed that there is a relationship between color intensity and polyphenol and anthocyanin content, that is, the more pigmented the shell, the higher will be the concentration of this compound in the grain. Moreover, the seed coat color of beans is frequently highly variable. The color variability is due to the existence of nine epistatic genes that are responsible for generating variations in the patterns of seed color. Table 3.5 shows the bean color using the color sense. *Phaseolus vulgaris* presented the highest a^* value (15.6 ± 1.5), which means that it is the species with the most intense red coloring. On the contrary, *Lablab Purpureus* presented the highest b^* value (17.0 ± 7.0) which means that it is the species with the most intense yellow coloring.

Table 3.5: Color representation, using the color sense

Sample	<i>Phaseolus lunatus</i>	<i>Lablab purpureus</i>	<i>Vigna unguiculata</i>	<i>Cajanus cajan</i>	<i>Phaseolus vulgaris</i>
Color intensity					

The dimensions of the seeds are part of the other important characteristics in the differentiation of one species from the other. The larger size was found in *Phaseolus lunatus* (both in length and width) and the smaller for *Cajanus cajan*.

3.2.4. Agronomic value of pulses

The development of Cabo Verde rural economy in recent years has been negative due to three consecutive years of drought, with irregular and insufficient rainfall (Costa, 2020). Included in the Sahel region, the archipelago experiences periods of drought characterized by the absence of rain, low rainfall or by its poor distribution over the rainy season (Neto *et al.*, 2020). Natural obstacles have historically been a challenge to Cabo Verde's rural

development. The contribution of agriculture to the rural economy decisively obeys to the favourable or adverse climatic conditions (Monteiro et al., 2020).

The evolution of the bean cultivated area follows a trend similar to the other rainfed crops, a consequence of being established alongside the maize crops (Fortes et al., 2020). The cultivation of the five bean species under study is of great importance, in terms of consumption/trade, for the food of rural households, occupying most of the agricultural areas of Santiago Island and dictating the food culture of this population (Kaufmann & Kubo, 2018). Despite the decreasing trend of cultivated areas in Cabo Verde, rainfed farming occupies a large portion, with 89.2% of the agricultural area occupied by maize and beans (MAA, 2017).

The different bean species are traditionally cultivated in association with maize, except for *Cajanus cajan* which is sown randomly (Barreto, 1996). This is a seasonal crop, directly dependent on the few months of rain (two to three months per year) concentrated between August to October (INMG, 2017). According to Temudo (2008), *Phaseolus vulgaris* is the most climatically demanding and the only bean species grown in the higher and cooler zones, where the precipitation is more regular and the air humidity is higher. On the other hand, *Lablab purpureus* is the most drought-resistant bean species (Temudo, 2008).

Altogether, the total production of traditional rainfed crops fluctuated from 2013 to 2017, decreasing from 12 008 tons (2013) to 700 tons (2014) and increasing to 9739 tons in 2016 (MAA, 2016). This variation pattern is also reflected in our results for beans, with an evident and strongly random character determined by the meteorological effects on crop production. It is worth noting that the negative trend of bean production contrasts with the average annual growth population rate of 12.1% over the last 10 years (2006-2016) (INE, 2018a). Moreover, it must be pointed out that *Cajanus cajan* and *Lablab purpureus* accounted for more than 50% of the total bean production (MAA, 2016).

The rainfed production in Cabo Verde still does not meet the needs of the population, ensuring only 10-15% of the national food consumption, thus forcing the huge importation of food supplies (MAP, 2002). This also applies to beans, as in addition to local production, a considerable consumption depends on importation of both canned and dried beans, mainly in the urban areas throughout the year, whereas the local production is consumed in the rural areas during a restricted period. Generally, the cultivated bean species are primarily used for

household self-support; however, in years of good harvests, beans are sold in the local markets (MAA, 2017).

3.2.5. Food security and pulses in Cabo Verde

Cabo Verde's history records cyclical famines, of which some stand out in the 1920s, 1940s and 1970s for decimating thousands of people (Lobo, 2015). More recently, Cabo Verde reported almost no harvests for the 2017-2018 agricultural season due to a severe drought, highlighting that 28 000 people (5.3%) are currently facing food insecurity (Data available at <http://www.west-africa-brief.org/content/en/partners-help-cabo-verde-cope-food-insecurity>).

Besides the environmental challenges to agriculture productivity, a recent study (Varela et al., 2020) revealed that Cabo Verde food production still falls short on meeting internal consumers' needs, posing a huge threat to food security and a high dependence on food imports (FAO & MDR, 2014). Despite the considerable progress made since independence (in 1975) in fighting poverty, Cabo Verde has not yet eradicated hunger and, according to FAO, about 20% of rural households suffer from food insecurity (DSSA, 2005).

Therefore, the lack of rain for the production of maize and beans leads to profound changes in the Cabo Verde food pattern, according to Silva (2005), the consumption of traditional products (maize and beans) that are the basis of the diet in Cabo Verde is being replaced by the consumption of 100% imported rice. This decrease in the production of maize and beans consequently leads to a decrease in grain consumption. Moreover, the purchase of food becomes more expensive. In this way, the food and nutritional security of families is negatively affected by reduction in the number of daily calories needed to keep their dietary needs balanced.

Grain legumes are a very important food crop in many parts of Africa, as they are a source of high-protein products, and have the advantage of fixing atmospheric nitrogen which enriches the soil thereby reducing the cost of fertilizer inputs, especially in nutrient poor soils (Vidigal et al., 2019). In Cabo Verde, rainfall scarcity and soil infertility, together with the arid territory pose a challenging scenario for agriculture production (Monteiro et al., 2020). It should be noted that the consumption of beans dates back to the first settlements of these islands, with the broad bean (*Vicia faba*) introduced by the Portuguese settlers and the African beans being brought to Cabo Verde from the western coasts of Africa, and from the New World, along with the slave trade (Torrão, 1995).

Maize has been a staple crop in Cabo Verde since its introduction from the Americas and still today, followed by legumes used for both food and fodder. Most of the beans traded in Santiago seem to be cultivated in Cabo Verde for several years, intercropping with potatoes and maize. Bean varieties and/or landraces adapted to Cabo Verde's severe conditions have been selected at smallholders' level. *Vigna unguiculata* (cowpea) is a drought-tolerant food crop, well adapted to a diverse range of climate and soil types, and widely cultivated throughout the tropics and subtropics (Appiah et al., 2011). In Africa, cowpea is mainly cultivated in West and Central Africa, with an annual production of 3 million tons, being also known as the poor man's meat (Onyenekwe et al., 2000). Also, *Lablab purpureus* (hyacinth bean) is among the so-called 'lost crops' but with potential to become an important crop species in the future due to its enhanced environmental tolerances when compared to other legumes (Maass et al., 2010; Robotham & Chapman, 2017). Considering its inherent environmental resilience, hyacinth bean in Cabo Verde can be seen, together with cowpea, as beans of prospected increased economic and social value in the future. Considering the historical cultivation of these species under the extreme environmental conditions of Cabo Verde, *ex situ* conservation measures should be considered to conserve these invaluable resources, as only 17 bean accessions (Supplementary Table S2) from Cabo Verde are held at germplasms (Genesys, 2020), namely: 14 of *Cajanus cajan*, two of *Phaseolus* (*Phaseolus lunatus* and *Phaseolus vulgaris*), and one of *Vigna unguiculata*. The adverse climatic conditions of this archipelago, with cyclical drought periods, surely drove acclimation processes and probably multiple landraces were locally developed in these islands. Interestingly, and despite the small number of edible grain legumes, more than 50% of the germplasm accessions reported from Cabo Verde are forage legumes (namely from the endemic-rich genus *Lotus*), revealing the interest of this family for pasture improvement (Duarte et al., 1996). *Ex situ* conservation of the plant genetic resources in Cabo Verde, especially concerning food species commonly used by the population, has been very limited, with few efforts to characterize, evaluate, and preserve this genetic heritage. Besides its great importance at nutritional and agronomic levels, the income raised from legume sales significantly contributes to food security at the household level (Ojiewo et al., 2015).

Legumes are a good source of protein and micronutrients and could hence be a good complement to starchy diets, where deficiency of protein is a concern (Abberton, 2010). Protein contents in legumes frequently range from 20 to 45%, which means a higher protein

content than most plant foods and twice the protein content of cereals (Leonard, 2012; FAO, 2016).

Conclusion

The present study involved an extensive research about the diversity of cultivated food legume species in Cabo Verde, focusing on the most consumed/traded pulses: *Cajanus cajan*, *Lablab purpureus*, *Phaseolus lunatus*, *Phaseolus vulgaris* and *Vigna unguiculata*. Through multiple approach – legumes diversity and local uses; chemical, nutritional and antioxidant evaluation; complemented with agro-economic analysis - results were discuss in the light of conservation and sustainable use of these legumes, and their potential contribution for food security in this archipelago. Notwithstanding, the results revealed that the studied bean species are good sources of minerals, proteins, phenols, and antioxidants, representing an invaluable potential to satisfy the nutritional needs of Cabo Verde populations. Beans represent about half of Cabo Verde's crop production which, together with maize, corresponds to about 90% of the total cropland. However, the adverse climatic conditions of this archipelago, characterised by the scarcity or irregularity of rainfall, causes a drastic decrease in local crop production. This translates into lack of food products, the need to import and the increase of prices, worsening the nutritional insecurity by reducing access to food. Under this scenario of food and socio-economic crisis, new strategies and investments must be implemented, such as the use of drought-resistant cultivars, new agricultural production techniques, and water saving and desalination systems, since most of the local population largely depends on domestic crop productivity for self-support.

Finally, this study highlights a limitation in *ex situ* conservation of the plant genetic resources of Cabo Verde. Thus, further field surveys are still needed, involving new efforts to enhance both *in situ* and *ex situ* conservation of these species, specifically to assess, catalogue, and preserve their genetic legacy that can be used in bean breeding programs.

In future works, it would be important to determine the nutritional composition of the leaves and phenolic compounds content present in seeds, to make a comparison between these two edible bean plant parts and contribute to their nutritional and nutraceutical valorization. It would still be relevant to perform a comparative nutritional study between green beans and

dried beans, so we can better unravel the differences between them, as they are both widely consumed. Finally, it would be interesting to carry out a study of the effect of heat treatment on beans minerals bioaccessibility.

CHAPTER V

References

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CHAPTER VI

Supplementary Data

Table S1. Eigenvalues, proportion of variability and mineral traits that contributed to the first four PCs (PCA) concerning the five most cultivated and traded food legume species in Cabo Verde.

Components	PC1	PC2	PC3	PC4
Eigenvalues	2.936	2.076	1.658	1.070
% Variance explained	29.36	20.76	16.58	10.70
% Cumulative variance	29.36	50.12	66.70	77.40
Coefficients of variance				
B	-0.503	0.098	-0.120	-0.201
Ca	-0.242	-0.559	0.091	-0.134
Cu	0.072	-0.099	-0.545	-0.216
Fe	0.059	-0.239	0.447	0.607
K	-0.396	-0.132	-0.237	0.158
Mg	-0.266	0.394	0.400	-0.020
Mn	0.347	0.485	0.112	-0.056
P	-0.420	0.215	0.009	0.147
S	-0.394	0.222	0.132	-0.485
Zn	-0.008	0.328	-0.484	0.492

Table S2. Accessions in worldwide genebanks of food legume species of Cabo Verde assessed through the Genesys Database (2020).

Species	Number of accessions	Biological status of accessions	Provenance (Island)
<i>Cajanus cajan</i>	9	Breeding/Research Material	Unknown
	5	Traditional cultivar/Landrace	Unknown
<i>Phaseolus lunatus</i>	1	Traditional cultivar/Landrace	Unknown
<i>Phaseolus vulgaris</i>	1	Traditional cultivar/Landrace	Unknown
<i>Vigna unguiculata</i>	1	Traditional cultivar/Landrace	Santiago

Interviewer's name _____			
Interview nr _____	Date _____	Place (market name) _____	
Name of interviewee _____			
Oral Consent .	Accepts our interview:	Yes ()	No()

QUESTIONNAIRE 1: IDENTIFICATION AND CHARACTERIZATION OF THE SELLER

1. Gender i) Male () ii) Female ()

2. How old are you? _____

- i) Child - 0 a 9 years () ii) Adolescent – 10 a 19 years () iii) Youth - 15 a 24 years ()
iv) Adult - 25 a 59 years () v) Old >60 anos

3. How many people live in your house? _____

4. What is your relationship with the head of the family?

- i) I am the head of the family () ii) Wife () iii) Son/daughter () iv) Other _____

5. What is your academic level?

- i) Primary (1 to 7 Class) () ii) Basic (8 to 10 Class) ()
iii) Medium (1 a 12) () iv) Graduate ()
v) Master () v) Other () _____

6. Civil Status

- i) Single () ii) Married () iv) Widower () v) Divorced ()
vii) Others () _____ vi) Separate ()

7. What is the main economic activity of your family (depends on what for survival)

- i) Agriculture () ii) Commerce () iii) Livestock () iv) Fishing () v) Other () _____

8- What products do you sell? _____

QUESTIONNAIRE 2: About beans

General Knowledge

1. How long time have you been selling beans?

- i) year ≤ 1 () ii) $1 < \text{Year} \leq 5$ () iii) $5 < \text{Years} \leq 10$ ()
vi) $10 < \text{Years} \leq 15$ () v) $15 < \text{Years} \leq 20$ () vi) Years > 20 ()

2. What is the origin of your customer?

- i) From the same municipality or town ()
ii) From the same islands ()
(iii) Other
Islands ()

3. What is the main reason why customers look for/buy beans?

- i) Customers enjoy my product ii) It's cheap
iii) It is very healthy iv) Another reason _____

4. Do you buy or produce? i) Buy () ii) Produce () (iii) both ()

5. From which area does the beans come ?

- i) Santiago () Region? _____
ii) Another Island of Cabo Verde () Which Island? _____
iii) Imported () From Where? _____
iv) Others
-

6. The sale of beans contribute to your family income?

- i) No ii) Not much iii) More or less
- iv) A lot v) Other_____

7. The income from the sale of beans serves to:

- i) Food () ii) Tuition fees () iii) Health () iv) House rent ()
- v) Other (), _____

8. How often customers buy/procure beans?

- i) Daily () ii) Weekly () iii) Variable/dependent (), specify _____
- iv) Other (), specify _____

9. What are the seasons where beans are sold the most?

- i) “Dry season/ January to July” () ii) “Rain season / August to December” () iii) Both ()

10. What are the main obstacles or problems you have had?
